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Experimental and Molecular Therapeutics

Abstract 1778: Characterization and anti-tumor functionality of a neuroblastoma-specific peptide, either free or conjugated to nanocarriers

Alice Bartolini, Monica Loi, Daniela D. Paolo, Laura Emionite, Angelina Sacchi, Flavio Curnis, Gianluca Bottoni, Michela Massollo, Cristina Gagliani, Silvia Bruno, Alessandro Gori, Renato Longhi, Michele Cilli, Carlo Tacchetti, Angelo Corti, Gianmario Sambuceti, Mirco Ponzoni, Serena Marchiò, and Fabio Pastorino

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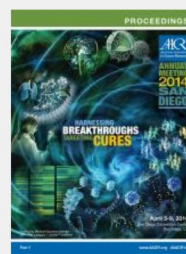
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Proceedings: AACR Annual Meeting 2014; April 5-9, 2014; San Diego, CA

Abstract

Introduction. The identification of peptide ligands specific for solid tumors is expected to provide targeting moieties to improve delivery and to decrease toxicity of chemotherapy. We have recently identified the peptide HSYWLRS as a specific ligand for neuroblastoma (NB), a childhood tumor mostly refractory to current therapies.

Experimental procedures. The capability of peptide HSYWLRS to recognize NB cells was evaluated by coupling Qdot fluorescent nanoparticles with HSYWLRS



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or its scrambled version (SCR). NB cell association and internalization of HSYWLRS-targeted liposomes were tested by FACS and confocal microscopy studies. We further evaluated a potential role of this peptide in perturbing tumor-stroma interactions and tumor growth. NB cell lines stably transfected with eGFP were mixed with endothelial cells in the presence of either SCR or HSYWLRS peptides. Cell morphology and reciprocal cellular interactions were evaluated by optical and fluorescence microscopy. We finally performed therapeutic experiments with mice orthotopically injected with luc-trasfected NB cells and treated with HSYWLRS-targeted, doxorubicin-loaded liposomes (HSYWLRS-SL[DXR]). Anti-tumor efficacy was evaluated by BLI imaging. In vivo imaging was also performed by injecting mice with a bolus of fluorodeoxyglucose during a list mode acquisition lasting one hour using a dedicated micro-PET system. After framing rate optimization, tumor glucose consumption was measured using Patlak graphical approach and normalizing the slope of regression line for serum glucose level.

Results. FACS analysis showed that HSYWLRS-Qdot and SCR-Qdot bound NB cells in a dose-dependent manner, however with different intensity, being HSYWLRS-Qdot the more potent. The binding of HSYWLRS-Qdot was efficiently inhibited by an excess of HSYWLRS, but not by control SCR peptide. In contrast, the binding of SCR-Qdot was not inhibited neither by an excess of SCR nor by HSYWLRS peptide, suggesting that the binding of SCR-Qdot is not specific. Again, the specific peptide-driven binding of HSYWLRS-SL to NB cells was inhibited by an excess of HSYWLRS peptide. In all cases, HSYWLRS specifically altered in vitro the interactions of NB cells with endothelium. Similarly, this peptide statistically decreased tumor take and growth when co-injected with tumor cells in the adrenal gland of nude mice.



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Preliminary in vivo results obtained by BLI and micro-PET devices indicated that HSYWLRS-SL[DXR] decrease tumor growth through a reduction of tumor glucose consumption, leading to an enhanced life span in treated mice.

Conclusion. Our findings demonstrate that HSYWLRS peptide recognizes NB cells and is functional in the design of nanocarriers with therapeutic efficacy paving the way to its clinical development.

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